DATA EVALUATION RECORD
FISH LIFE-CYCLE TOXICITY TEST
§72-5

1. **CHEMICAL:** Flufenacet   PC Code No.: 121903

2. **TEST MATERIAL:** FOE 5043 Technical   Purity: 95.6%

3. **CITATION:**

Author:	Hall, A.T., and C.V. Lam
Title:	Fathead Minnow (*Pimephales promelas*) Fish Life Cycle Test with Flufenacet (FOE 5043 Technical)
Study Completion Date:	October 28, 2002
Laboratory:	Bayer Corporation 17745 South Metcalf Stilwell, KS 66085-9104
Sponsor:	Bayer Corporation P.O. Box 4913 Kansas City, MO 64120-0013
Laboratory Report ID:	F3851201 (Bayer Report No. 109767)
MRID No.:	45796114
DP Barcode:	D288186

4. **REVIEWED BY:** Rebecca Bryan, Staff Scientist, Dynamac Corporation

   Signature: Date: 12/29/03

   APPROVED BY: Christie E. Padova, Staff Scientist, Dynamac Corporation

   Signature: Date: 12/29/03

5. **APPROVED BY:** Jennifer Leyhe, Biologist, OPP/EFED/ERB - V

   Signature: Date: 10/23/06

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     Signature: *Rebecca Bryan*   **Date:** 12/29/03

     APPROVED BY: Christie E. Padova, Staff Scientist, Dynamac Corporation

     Signature: *C. E. Padova*   **Date:** 12/29/03

5. **APPROVED BY:** Alan Vaughan, OPP/EFED/ERB - V

     Signature: **Date:**

6. <u>STUDY PARAMETERS</u>:

Scientific Name of Test Organism:	*Pimephales promelas*
Age of Test Organism:	<24 hours old (F_0 generation)
Definitive Test Duration:	289 Days (9.5 months)
Study Method:	Flow-through
Type of Concentrations:	Mean-measured

7. <u>CONCLUSIONS</u>:

The 9.5-month chronic toxicity of FOE 5043 Technical (flufenacet) to the full life stage of Fathead Minnow (*Pimephales promelas*) was studied under flow-through conditions. Fertilized eggs (approximately 200 embryos/treatment, <24 hours old) were exposed to the test material at nominal concentrations of 0 (negative and solvent controls), 0.087, 0.175, 0.35, 0.70, and 1.4 ppm. Mean-measured concentrations were <0.009 (<LOQ, controls), 0.075, 0.138, 0.274, 0.600, and 1.211 ppm a.i.

Following hatching on Day 6, alevins were reduced to 100 per treatment level. On Day 36, the juveniles were again reduced to 60 per treatment level. On Day 146, the aquaria were equipped to accommodate spawning with one male and two females assigned to each spawning aquaria. Beginning on Day 254, embryos of the F_1-generation were isolated and exposed under identical test conditions as those described for the parental generation until 35 days post-hatch.

<u>F_0-generation</u>: No treatment-related effects were observed on embryo survival, hatching success, larval, juvenile, or adult survival of the F_0 generation. In addition, no significant treatment-related effects were observed on egg production at any test level.

Growth was affected by treatment with flufenacet: the most sensitive endpoint was adult male weight at test termination (Day 254), with a NOEC of 0.075 ppm a.i.

<u>F_1-generation</u>: Growth was also affected at the 1.211 ppm a.i. level 4-week post-hatch in the filial generation. No other treatment-related effects were observed on hatching success, or egg or alevin/fry survival.

Based on the male test termination weights of the F_0 generation, the NOEC and LOEC are 0.075 and 0.138 ppm a.i., respectively.

This study is classified as SUPPLEMENTAL. This study did not fulfill the guideline requirements for a fish life-cycle toxicity test because the F_1 generation was only maintained for 4 weeks post-hatch. This study is scientifically valid, and although results do not meet guideline requirements; the information may be useful in a risk assessment.

Results Synopsis:

Biological Endpoint	NOEC (ppm a.i.)	LOEC (ppm a.i.)
F_0 hatching success	1.211	>1.211
F_0 egg survival	1.211	>1.211
F_0 4-week survival (Day 36)	1.211	>1.211
F_0 4-week length (Day 36)	1.211	>1.211
F_0 8-week length (Day 63)	0.600	1.211
F_0 20-week survival (Day 146)	1.211	>1.211
F_0 33-week survival (Day 237)	1.211	>1.211
F_0 test termination survival (Day 254)	1.211	>1.211
F_0 test termination length (Males)	0.274	0.600
F_0 test termination length (Females)	1.211	>1.211
F_0 test termination weight (Males)	0.075	0.138
F_0 test termination weight (Females)	1.211	>1.211
F_0 egg production, Days 148 - 237	1.211	>1.211
F_0 egg production, Days 148 - 254	1.211	>1.211
F_0 egg viability, Days 148-237	1.211	>1.211
F_0 # of spawns/female	1.211	>1.211
F_0 # of eggs/female	1.211	>1.211
F_0 # of eggs/spawn	1.211	>1.211
F_1 hatching success	1.211	>1.211
F_1 egg survival	1.211	>1.211

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Biological Endpoint	NOEC (ppm a.i.)	LOEC (ppm a.i.)
F_1 4-week survival (Day 35)	1.211	>1.211
F_1 4-week length (Day 35)	0.600	1.211
F_1 4-week weight (Day 35)	0.600	1.211

NOEC: 0.075 ppm a.i.
LOEC: 0.138 ppm a.i.
Endpoint(s) affected: F_0 and F_1 generations: growth.
Most sensitive endpoint: Adult male (F_0) growth at study termination.

8. ADEQUACY OF THE STUDY:

A. Classification: Supplemental

B. Rationale: Because the F_1 generation was only maintained for 4 weeks post-hatch (instead of the required 8 weeks), this study does not satisfy guideline requirements for a fish life-cycle toxicity test (§72-5). This study is scientifically valid, and provides supplemental data on the toxicity of FOE 5043 Technical (flufenacet) to the life cycle of fathead minnow.

C. Repairability: This study may be upgraded to Core status if data are provided to support that assumptions of no adverse effects on the survival, appearance, or growth of second-generation larvae at the ≤0.600 ppm a.i. test levels would have been maintained, had the fish been observed up to 8 weeks post-hatch.

9. GUIDELINE DEVIATIONS:

1.   Survival of F_0-generation fish was not recorded 8 weeks following hatch.

2. F_1-generation fish were maintained for only 4 weeks, instead of the required 8 weeks.

3.   The test vessels were aerated during the test (started on day 92). Thereafter, on occasion, DO levels fell <60% saturation (on Days 104, 111, 150, 153, 174, 185, 197, 213, 248, 251, and 249 in the extended growth/spawning chambers and on Days 150 and 240 in the growth chambers). Aeration did not appear to have an affect on test concentration levels.

4.   The flow-splitting accuracy was not reported.

10. SUBMISSION PURPOSE: This study was submitted to provide data on the toxicity of Flufenacet to the life cycle of fathead minnows for the purposes of chemical registration.

11. MATERIALS AND METHODS:

A. Test Organisms

Guideline Criteria	Reported Information
Species Prefer Sheepshead minnow (*Cyprinodon variegatus*) or Fathead minnow (*Pimephales promelas*).	Fathead minnow (*Pimephales promelas*)
Source and Acclimation	Embryos were obtained from in-house laboratory breeding cultures. The culture dilution water was from the same source as the dilution water used for testing. Pre-test mortality was not reported.
Age at beginning of test Embryos, 2 to 24 hours old	Embryos, <24 hours old
Feeding Fish should be fed at least twice daily and should not be fed for at least 24 hours prior to test termination.	Newly hatched fry were fed brine shrimp nauplii one to two times daily during testing. After day 7, a ground trout chow or Tetra-Min was supplemented. The amount of food increased during the study

Guideline Criteria	Reported Information
	to compensate for growth. The fish were not fed for 24 hours prior to test termination.
Embryo Exposure (4 to 5 Days) Embryos (≤24 hours old) from at least 3 separate spawns should be randomly distributed to embryo cups. A minimum of 50 embryos (≤24 hrs old) per replicate cup, 4 cups per treatment should be used. Parameters measured: · Survival of embryos · Time required to hatch · Hatching success · Survival of fry for 4 weeks Dead and fungused embryos should be counted and removed daily.	Days 0-6 Embryos (< 24 hours old) were obtained from brood stock and randomly assigned into embryo incubation cups. Each cup contained 50 embryos, with two cups per replicate and two replicate aquaria per treatment level (total of 200 embryos per treatment). Three additional egg cups of 50 eggs were used to determine viability. Parameters measured: · Survival of embryos · Time required to hatch · Hatching success · Viability (Day 2) · Survival of fry for 4 weeks Mortality was determined daily, and dead and fugused embryos were removed.
Larval-Juvenile Exposure (From Hatch to 8 Weeks) After hatching, each group of larvae is randomly reduced to a minimum of 25 fish and released in replicate larval growth chambers. The random selection must include any fish that are lethargic or deformed. Parameters measured: · Fish survival (determined by counting the number of live fish in	Days 6-63 (hatch to approximately 8 weeks) When the hatching period was completed, larvae were reduced to 25 per replicate, with four replicates per treatment (100 embryos per treatment), and the larvae were transferred from the incubation cups to the larval growth chambers in the corresponding aquarium. On Day 36, fish were thinned to 15 fish per growth chamber and transferred to extended growth chambers (30 fish per extended growth chamber, 60 fish per treatment).

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Guideline Criteria	Reported Information
each replicate growth chamber weekly). · Total lengths (mm) of all fish at 4 and 8 weeks after hatching.	Parameters measured: · Survival of fry/juvenile fish (weekly). · Lengths (mm) of all surviving fish at 34 and 63 Days (approximately 4 and 8 weeks).
Juvenile-Adult Exposure (From 8 weeks posthatch to the end of the spawning phase [32-40 weeks]) At 20-24 weeks after hatching, mature fish are placed in a spawning tank of the same concentration (4 males and 4 females randomly chosen and assigned). The spawning tank is divided into 4 individual spawning chambers with appropriate spawning substrates. The substrates are examined daily and embryos removed, counted, and recorded separately for each pair. For fathead minnow, adult exposure should be terminated when no spawning occurs for one week. For sheepshead minnow, testing should be terminated after spawning is observed for 2 weeks.	Days 63 to 254 On Day 146, spawning substrates were added to the aquaria and one male and two females assigned to each spawning area. There were four spawning areas per aquaria (two replicate aquaria per treatment). The spawning substrates are examined daily and embryos removed, counted, and examined for viability (number of viable, non-viable, and popped eggs). Adult exposure was terminated on day 254. Parameters measured: · Survival of adult fish · Spawning frequency · Number of eggs deposited · Lengths (mm) and wet weight (g) of all surviving fish at 254 days
Second Generation Embryo Exposure (4 to 5 days) 50 embryos from each conc. level are randomly selected and transferred to incubation cups for hatch. Use the same test procedures as those for parental generation. Embryos not selected are discarded.	Days 0-6 (254-260) In each aquarium, ≤50 embryos per replicate aquarium (100 per level) were incubated for the early life-stage test, and an additional ≤35 embryos per replicate were incubated for hatching success (only). The same test procedures as those employed for the parental generation were used. The embryos were observed daily.

Guideline Criteria	Reported Information
Second Generation Larval-Juvenile Exposure (From Hatch to 4-8 weeks) After hatching, 25 larvae are released in each growth chambers (2 chambers per treatment).	Days 7-35 (261-289) After hatching, the hatching success study was terminated, and early life-stage fry were reduced to 25 alevins per replicate aquarium (50 per level).
Each group of 2nd generation fish is terminated 8 weeks after hatching.	Each group of F_1-generation fish was terminated 4 weeks after hatching.
Fish are blotted, weighed, and measured before being discarded.	Fish were weighed and measured for length.

Comments: Three replicate test vessels were used to check the viability of the eggs used to initiate the fish life-cycle test (p. 19). The mean percent embryo viability on Day 2 was 92%, which indicated good viability of the eggs from the stock cultures.

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B. Test System

Guideline Criteria	Reported Information
Test Water Sheepshead Minnow 1. Natural seawater (sterilized and filtered) or a commercial mixture. 2. Natural seawater with a salinity of ≥ 15 parts per thousand (weekly range of salinity <6% and monthly pH range <0.8 pH units). Fathead Minnow 1. Reconstituted water or water from unpolluted well or spring (sterilized and tested for pollutants). 2. Hardness of 40 to 48 mg/L as $CaCO_3$ and pH of 7.2 to 7.6.	N/A 1. The dilution water was produced by blending spring water that had been filtered and UV-sterilized with city water that had been de-chlorinated, filtered, de-mineralized, and treated by reverse osmosis. The resultant dilution water (soft) was stored in polypropylene or PVC holding tanks and intensely aerated and UV-sterilized prior to use. 2. Hardness of 36 to 66 mg/L as $CaCO_3$ and pH of 7.3 to 7.9.
Test Temperature Sheepshead: 30°C. Fathead: 25°C and should not remain outside the range of 24 to 26°C for more than 48 hours.	N/A Range of 23.5 to 26.7°C. The temperature fluctuated outside of 25 ± 1°C on five occasions, for a total duration of 16 hours (p. 18).
Photoperiod 16-hour light/8-hour dark. Light intensity of 10-100 lumens at water surface.	The photo-period followed the natural photo-period times in Evansville, Indiana, with 30-minute transition periods (Table 2, p. 31). The photo-periods varied from 10 hours and 30 minutes to 15 hours and

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Guideline Criteria	Reported Information
	45 minutes of light during the study. The light intensity average was 69.7 foot-candles.
Dosing Apparatus 1. Intermittent flow proportional diluters or continuous flow serial diluters. 2. A minimum of 5 toxicant concentrations with a dilution factor ≤ 0.5. 3. One control should be used.	1. Mount and Brungs proportional diluter system. 2. Five toxicant concentrations with a dilution factor of 0.5. 3. A negative control and a solvent control was used.
Toxicant Mixing 1. Mixing chamber recommended but not required. 2. Test solution completely mixed before introduction into the test system (aeration should not be used for mixing). 3. Flow splitting accuracy must be within 10% and periodically checked.	1. No mixing chamber was reported. 2. Yes 3. The flow-splitting accuracy was observed one to two times daily during the test.
Exposure System/Test Vessels Exposure tanks should be all glass or glass with a plastic or stainless steel frame (30.5 x 30.5 x 91.4 cm or 30.5 x 30.5 x 61 cm for fathead, and 45 x 90 x 26 cm for sheepshead). Larval chambers should have glass bottoms and drains that allow water to be drawn down to 3 cm. Test water depth in adult tanks and larval chambers should be a minimum of 15 cm.	Glass exposure aquaria that measured 39.7 L (30.5 x 52 x 30.5 cm) were used as extended growth chambers. These chambers were divided into 4 separate spawning areas using perforated stainless steel partitions (20 gauge, 0.156 in diameter holes). The larval growth chambers measured 12.5 x 21.5 x 30.5 cm with a fill volume of 6.8 L.

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Guideline Criteria	Reported Information
	It was not specified if test chambers had drains to allow for water level reduction.
Embryo and Fry Chambers 120 mL glass jars with bottoms replaced with 40 mesh stainless steel or nylon screen. Chambers can be oscillated vertically using rocker arm apparatus (2 rpm motor) or placed in separate chambers with self-starting siphons.	The egg incubation cups were glass jars (7 cm diameter, 11.5 cm height, and 440 mL volume) with 20 mesh nytex screen bottoms. The egg cups were placed in the growth chamber and oscillated in the test solution using a rocker arm apparatus.
Flow Rate Flow rates to adult tanks or larval chambers should provide 90% replacement in 8-12 hours, and maintain DO at above 75% of saturation. The toxicant level cannot drop below 20% with fish in the tank.	During the test, the flow rate was at least 4 volume additions per day. The dissolved oxygen levels fell below the 75% saturation.
Aeration Dilution water should be aerated to insure dissolved oxygen concentrations at or near 100% saturation. Test tanks and embryo chambers should not be aerated.	The test vessels were aerated starting on Day 92 of the test.

C. Chemical System

Guideline Criteria	Reported Information
Nominal Concentrations Minimum of 5 concentrations and a control, all replicated; plus solvent control if appropriate. Toxicant conc. must be measured in one tank at each toxicant level every week.	0 (negative and solvent controls), 0.087, 0.175, 0.35, 0.70, and 1.4 ppm. Toxicant concentrations were measured weekly from alternating replicate aquaria in each test group.
Other Variables	

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Guideline Criteria	Reported Information
1. DO must be measured at each conc. at least once a week. 2. Test water temp. must be recorded continuously. 3. Freshwater: A control and one conc. must be analyzed weekly for pH, alkalinity, hardness, and conductance. Natural seawater: must maintain a constant salinity and not fluctuate more than 6% weekly; monthly pH range <0.8 pH units.	1. DO was measured weekly in all replicate aquariums (that contained test organisms), and as needed to monitor recoveries during low DO periods. 2. Temperature was measured in hourly in one centrally located test aquarium. Temperature was also recorded daily. 3. The pH, hardness, alkalinity, and conductivity were measured weekly in the controls, low, middle, and high test concentrations.
Solvents Should not exceed 0.1 ml/L in a flow-through system. Acceptable solvents are: dimethylformamide, triethylene glycol, methanol, acetone, ethanol.	Acetone, 0.1 mL/L

Comments: The study authors reported that due to the presence of acetone in the test systems, maintaining DO concentrations throughout the test was difficult (p. 19). Extensive cleaning (multiple times/week) in addition to aeration (starting on Day 92) were used. On occasion, DO levels fell <60% saturation (on Days 104, 111, 150, 153, 174, 185, 197, 213, 248, 251, and 249 in the extended growth/spawning chambers and on Days 150 and 240 in the growth chambers).

12. **REPORTED RESULTS:**

A. General Results

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes
<u>Data Endpoints</u> must include: · survival of P and F_1 embryos, time required to hatch, and hatching success; · survival and total length of P fish at 4 and 8 weeks after hatching; · weights and lengths of F_1 fish at 8 weeks; · incidence of pathological or histological effects; and · observations of other effects or clinical signs.	<u>Data Endpoints</u> included: · survival of F_0 and F_1 embryos, time required to hatch, and hatching success; · survival of F_0 fish at 4, 20, 33 (just prior to Ick), and 35 weeks (Day 254, study termination) after hatching; · total length of F_0 fish at 4 and 8 weeks after hatching; · total length, and wet weight of surviving F_0 fish at 254 Days (gender specific); · no. viable, non-viable, popped, and total eggs of F_0 spawning groups; · no. eggs/spawn, eggs/female, and spawns/female of F_0 spawning groups; · wet weights and lengths of F_1 fish at 4 weeks after hatching; · incidence of pathological or histological effects; · observation of other effects or clinical signs
Raw data included?	Yes

F₀ Results:

Nominal Conc. (ppm)	Mean Measured Conc. (ppm a.i.)	% Hatch[1]	4-Week Post-Hatch % Survival	20-Week Post-Hatch % Survival	33-Week Post-Hatch % Survival	35-Week (Day 254) Post-Hatch % Survival
Negative Control	<0.009	96.1	90	100	100	92
Solvent control	<0.009	94.7	86	98	92	79
0.087	0.075	97.9	91	93	96	83
0.175	0.138	98.9	81	98	75	71
0.35	0.274	95.6	84	95	96	96
0.70	0.600	93.9	93	98	83	83
1.4	1.211	94.3	81	98	92	83

Data obtained from Tables 6, 8, 9, and 12, pp. 42, 44, 45, and 48, respectively.
[1] Corrected for viability.

Mean Measured Conc. (ppm a.i.)	Mean Total Length (mm)				Wet Weight (g)	
	4 Weeks Post-Hatch	8 Weeks Post-Hatch	Day 254 (Test Termination)		Day 254 (Test Termination)	
			♂	♀	♂	♀
Negative Control	19.6	37.2	57.8	44.6	4.8922	2.1858
Solvent control	20.7	35.7	57.8	46.4	5.8149	2.4203
0.075	19.7	37.2	58.6	46.3	5.0761	2.0871
0.138	19.9	38.2	57.2	47.8	4.6736	2.3539
0.274	19.9	35.8	57.0	45.9	4.2453**	2.0255
0.600	19.5	35.7	52.6	45.9	3.9384**	2.0047
1.211	19.6	31.6*	54.0	45.3	3.6052**	1.8106*

Data obtained from Tables 10-11 and 13-14, pp. 46-47 and 49-50, respectively.
* Significantly different from the pooled control (p<0.05).
**Significantly different from the solvent control (p<0.05).

Mean Measured Conc. (ppm a.i.)	Total Eggs Days 148-254	Number of Eggs/ Spawn	Number of Eggs/ Female	Number of Spawns/Female	Mean % Viable Eggs
Negative Control	4919	129	2102	16	88.2
Solvent control	5431	179	2303	13	87.5
0.075	5862	143	2455	17	87.9
0.138	5812	181	2671	13	87.8
0.274	4926	170	2134	12	89.5
0.600	4507	132	2096	14	83.3
1.211	2250*	108	1056	8	77.2*

Data derived from Tables 15-17, pp. 51-53.
* Significantly different from the pooled control (p<0.05).

Toxicity Observations: No treatment-related effects were observed on embryo survival,   hatching suc
incubation. By 4 weeks post-hatch, fry survival ranged from 81 to 93% for all treatment
groups and controls. By 20 week post-hatch (Day 146), fry survival ranged from 93 to 100%
for all treatment groups and controls. By test termination (Day 254), fry survival ranged
from 71 to 96% for all treatment and control groups.

Growth (length and weight) of the F_0 generation was affected by treatment with Flufenacet.
A statistically-significant reduction in total length was observed at 8 weeks post-hatch
between the pooled controls and 1.211 ppm a.i. test group (36.4 mm versus 31.6 mm; Table
1, p. 47). In addition, at study termination (Day 254), statistically-significant reductions in
wet weight (compared to solvent control) were observed in males from the 0.274, 0.600, and
1.211 ppm a.i. test groups, and in females from the 1.211 ppm a.i. test group (Tables 13 and
14, pp. 49-50).

The number of total eggs produced on Days 148-254 were significantly different from the
pooled controls at the 1.211 ppm a.i. treatment level (5175 versus 2250; Table 15, p. 51).
However, no significant difference was detected in egg production from Days 148 to 237. In
addition, a statistically-significant reduction in the mean percent of viable eggs was observed
between the pooled controls and 1.211 ppm a.i. treatment group (87.8 versus 77.2%; Table
16, p. 52).

No treatment-related signs of toxicity were observed (pp. 20-22).

F_1 Results:

Mean Measured Concentration (ppb a.i.)	% Hatch	4-Week Post-Hatch % Survival	4-Week Post-Hatch Length (mm)	4-Week Post-Hatch Wet Weight (g)
Negative Control	98.1	93	22.8	0.2202
Solvent control	94.0	81	22.1	0.2155
0.075	91.0	90	22.3	0.2153
0.138	93.1	93	22.7	0.2142
0.274	97.5	93	22.1	0.2020
0.600	91.4	91	21.9	0.2024
1.211	77.4*	87	20.3*	0.1591*

Data obtained from Tables 18 and 20-21, pp. 54 and 56-59.
* Significantly different from the pooled control (p<0.05).

Toxicity Observations: In the F_1 generation, statistically-significant reductions compared to pooled controls were observed in percent hatch, and 4-week post-hatch total lengths and wet weights at the 1.211 ppm a.i. treatment level. Hatching began on Day 2 for all test and control groups, with no treatment-related differences in the number of hatched larvae. Hatching was completed on Day 7 for all groups.

No treatment-related signs of toxicity were observed in surviving fry during the 35 day observation period (pp. 25-26).

B. Reported Statistical Results

Data obtained for the F_0 generation that were statistically analyzed included (1) hatching success, (2) survival, (3) total length at 34 and 63 days post-hatch, (4) total length and wet weight on Day 254 (study termination), (6) spawning frequency (eggs/spawn, spawns/female, and eggs/female). Data obtained for the F_1 generation that were statistically analyzed included (1) hatching success, (2) survival, (3) and total length and wet weight at 35 days post-hatch.

The negative and solvent control data was compared using a t-test. If there was a significant difference the solvent control was used for comparisons (only F_0 Day 254 male weight data). The controls were pooled for data where the controls were not significantly different. The chi-square test was used to test for normality and the Bartlett's test was used to test for homogeneity to variance. The ANOVA followed by the Dunnett's test (control groups not pooled), the Bonferroni test (control groups pooled), or a nonparametric analysis (normality or homogeneity of variance assumptions not met) were used to evaluate difference between

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treatment and the control means (p≤0.05). The statistical analysis was performed using a computer program (TOXSTAT).

The no observed effect concentration (NOEC) is the highest test concentration causing no adverse effects. The lowest observed effect concentration (LOEC) is the lowest test concentration causing adverse effects. The maximum acceptable toxicant concentration (MATC) was calculated as the geometric mean of the NOEC and the LOEC.

Biological Endpoint	NOEC (ppm a.i.)	LOEC (ppm a.i.)
F_0 hatching success	1.211	>1.211
F_0 4-week survival (Day 36)	1.211	>1.211
F_0 4-week length (Day 34)	1.211	>1.211
F_0 8-week survival (Day 63)	Not determined	Not determined
F_0 8-week length (Day 63)	0.600	1.211
F_0 8-week weight	Not determined	Not determined
F_0 20-week survival (Day 146)	1.211	>1.211
F_0 test termination survival	1.211	>1.211
F_0 test termination length (Males)	1.211	>1.211
F_0 test termination length (Females)	1.211	>1.211
F_0 test termination weight (Males)	0.138	0.274
F_0 test termination weight (Females)	0.600	1.211
F_0 egg production, Days 148 - 237	1.211	>1.211
F_0 egg production, Days 148 - 254	0.600	1.211
F_0 egg viability, Days 148-237	0.600	1.211
F_0 # of spawns/female	1.211	>1.211
F_0 # of eggs/female	1.211	>1.211
F_0 # of eggs/spawn	1.211	>1.211
F_1 hatching success	0.600	1.211

Biological Endpoint	NOEC (ppm a.i.)	LOEC (ppm a.i.)
F_1 4-week survival (Day 35)	1.211	>1.211
F_1 4-week length (Day 35)	0.600	1.211
F_1 4-week weight (Day 35)	0.600	1.211
F_1 8-week survival	Not determined	Not determined
F_1 8-week length	Not determined	Not determined
F_1 8-week weight	Not determined	Not determined

NOEC: 0.138 ppm a.i.
LOEC: 0.274 ppm a.i.
MATC: 0.194 ppm a.i.
Endpoint(s) affected: F_0 generation: growth, egg production, and egg viability.
F_1 generation: growth.
Most sensitive endpoint: Adult male (F_0) growth at study termination.

13. **REVIEWER'S STATISTICAL RESULTS:**

Endpoints analyzed for the parental generation (F_0) included % hatch, egg and alevin survival, Day 36 survival, Day 146 survival, adult survival (Days 237 and 254), length (Days 34 and 63), adult male and female length and weight, egg production (Days 148-237 and 148-254), egg viability (Days 148-237), no. eggs/female, no. eggs/spawn, and no. spawns/female. Endpoints analyzed for the first filial generation (F_1) included % hatch, egg and alevin survival, Day 35 survival, length, and weight. With the exception of adult survival (Day 237), no. spawns/female, F1 % hatch, and F1 egg and alevin survival, data for all analyzed endpoints satisfied the assumptions of ANOVA (i.e., normal distribution and variance homogeneity), so the NOEC and LOEC for these endpoints were determined using ANOVA, followed by Bonferroni's test or William's test (if necessary). Data which did not satisfy the assumptions of ANOVA were analyzed using the non-parametric Kruskal-Wallis test. For all endpoints, the solvent control was compared to the negative control using a Student's t-test and if no difference was detected, the two were pooled for comparison to treatment; if a difference was detected, the treatment groups were compared to the solvent control. These analyses were conducted using TOXSTAT statistical software using mean-measured concentrations.

Biological Endpoint	NOEC (ppm a.i.)	LOEC (ppm a.i.)
F_0 hatching success	1.211	>1.211
F_0 egg survival	1.211	>1.211
F_0 4-week survival (Day 36)	1.211	>1.211
F_0 4-week length (Day 36)	1.211	>1.211
F_0 8-week length (Day 63)	0.600	1.211
F_0 20-week survival (Day 146)	1.211	>1.211
F_0 33-week survival (Day 237)	1.211	>1.211
F_0 test termination survival (Day 254)	1.211	>1.211
F_0 test termination length (Males)	0.274	0.600
F_0 test termination length (Females)	1.211	>1.211
F_0 test termination weight (Males)	0.075	0.138
F_0 test termination weight (Females)	1.211	>1.211
F_0 egg production, Days 148 - 237	1.211	>1.211
F_0 egg production, Days 148 - 254	1.211	>1.211
F_0 egg viability, Days 148-237	1.211	>1.211
F_0 # of spawns/female	1.211	>1.211
F_0 # of eggs/female	1.211	>1.211
F_0 # of eggs/spawn	1.211	>1.211
F_1 hatching success	1.211	>1.211
F_1 egg survival	1.211	>1.211
F_1 4-week survival (Day 35)	1.211	>1.211
F_1 4-week length (Day 35)	0.600	1.211
F_1 4-week weight (Day 35)	0.600	1.211

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NOEC: 0.075 ppm a.i.
LOEC: 0.138 ppm a.i.
Endpoint(s) affected: F_0 and F_1 generations: growth.
Most sensitive endpoint: Adult male (F_0) growth at study termination.

14. REVIEWER'S COMMENTS:

The reviewer's conclusion regarding the most sensitive endpoint (F_0 adult male weight) was identical to the study authors'; however, the reviewer's NOEC and LOEC values for this and other endpoints (i.e., F_0 adult female weight, F_0 adult male length, egg production for Days 148-254, egg viability for Days 148-237, F_1 % hatch, and F_1 egg and alevin survival) differed from the those of the study authors. The differences between the reviewer's and study authors' toxicity values are due to the different statistical methods used to estimate these values. The reviewer's conclusions are reported in the Executive Summary and Conclusions sections; the NOEC and LOEC for this study are 0.075 and 0.138 ppm, respectively.

The fish in all treatment groups and controls were treated for the disease Ichthyophthiriasis on Day 240 with Ick Guard (p. 16). The total egg production was also analyzed from the beginning of the spawning phase to the day prior to the Ick symptoms. The study author reports that it had minimal impact on the validity of the results.

The test chambers were cleaned as often as necessary to eliminate excess fecal material, food and microbial growth.

The diluter malfunctioned periodically during the test, which is not considered unusual for long-term chronic tests (See Appendix 2 footnotes, p. 103).

The dissolved oxygen fell below 60% saturation during the test (range of 27 to 99% saturation). Aeration was added to test chambers on day 92 to maintain DO levels.

It was not specified if test chambers had drains to allow for water level reduction.

This study was performed according to U.S. EPA (FIFRA) Good Laboratory Practice Standards (40 CFR 160) with the exception of the dilution water analysis. A Quality Assurance Statement was provided.

15. REFERENCES:

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16. OUTPUT OF REVIEWER'S STATISTICAL VERIFICATION:

F0 % hatch
File: 6114h   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	66.857	13.371	0.776
Within (Error)	22	379.250	17.239	
Total	27	446.107		

Critical F value = 2.66 (0.05,5,22)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

F0 % hatch
File: 6114h   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	88.250	88.250		
2	0.075	90.750	90.750	-0.983	
3	0.138	91.500	91.500	-1.278	
4	0.274	88.500	88.500	-0.098	
5	0.600	87.000	87.000	0.492	
6	1.211	87.500	87.500	0.295	

Bonferroni T table value = 2.51   (1 Tailed Value, P=0.05, df=22,5)

F0 % hatch
File: 6114h   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	8			

22

2	0.075	4	6.379	7.2	-2.500
3	0.138	4	6.379	7.2	-3.250
4	0.274	4	6.379	7.2	-0.250
5	0.600	4	6.379	7.2	1.250
6	1.211	4	6.379	7.2	0.750

F0 % hatch
File: 6114h   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP IDENTIFICATION		N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	8	88.250	88.250	89.688
2	0.075	4	90.750	90.750	89.688
3	0.138	4	91.500	91.500	89.688
4	0.274	4	88.500	88.500	88.500
5	0.600	4	87.000	87.000	87.250
6	1.211	4	87.500	87.500	87.250

F0 % hatch
File: 6114h   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	89.688				
0.075	89.688	0.565		1.72	k= 1, v=22
0.138	89.688	0.565		1.80	k= 2, v=22
0.274	88.500	0.098		1.83	k= 3, v=22
0.600	87.250	0.393		1.84	k= 4, v=22
1.211	87.250	0.393		1.85	k= 5, v=22

s = 4.152
Note: df used for table values are approximate when v > 20.

F0 egg and alevin survival
File: 6114s   Transform: NO TRANSFORMATION

ANOVA TABLE

23

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SOURCE	DF	SS	MS	F
Between	5	61.357	12.271	0.541
Within (Error)	22	498.750	22.670	
Total	27	560.107		

Critical F value = 2.66 (0.05,5,22)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

F0 egg and alevin survival
File: 6114s   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	85.000	85.000		
2	0.075	86.250	86.250	-0.429	
3	0.138	88.500	88.500	-1.200	
4	0.274	87.500	87.500	-0.857	
5	0.600	84.000	84.000	0.343	
6	1.211	87.000	87.000	-0.686	

Bonferroni T table value = 2.51   (1 Tailed Value, P=0.05, df=22,5)

F0 egg and alevin survival
File: 6114s   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	8			
2	0.075	4	7.315	8.6	-1.250
3	0.138	4	7.315	8.6	-3.500
4	0.274	4	7.315	8.6	-2.500
5	0.600	4	7.315	8.6	1.000
6	1.211	4	7.315	8.6	-2.000

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F0 egg and alevin survival
File: 6114s   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1 GRPS 1&2 POOLED	8	85.000	85.000	85.000
2 0.075	4	86.250	86.250	86.250
3 0.138	4	88.500	88.500	86.667
4 0.274	4	87.500	87.500	86.667
5 0.600	4	84.000	84.000	86.667
6 1.211	4	87.000	87.000	87.000

F0 egg and alevin survival
File: 6114s   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	85.000				
0.075	86.250	0.429		1.72	k= 1, v=22
0.138	86.667	0.572		1.80	k= 2, v=22
0.274	86.667	0.572		1.83	k= 3, v=22
0.600	86.667	0.572		1.84	k= 4, v=22
1.211	87.000	0.686		1.85	k= 5, v=22
----

s = 4.761
Note: df used for table values are approximate when v > 20.

d36 survival
File: 6114d36   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	534.857	106.971	2.884
Within (Error)	22	816.000	37.091	

Total   27   1350.857

Critical F value = 2.66 (0.05,5,22)
Since F > Critical F REJECT Ho:All groups equal

d36 survival
File: 6114d36   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

| | | TRANSFORMED | MEAN CALCULATED IN | | |
GROUP	IDENTIFICATION	MEAN	ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	88.000	88.000		
2	0.075	91.000	91.000	-0.804	
3	0.138	81.000	81.000	1.877	
4	0.274	84.000	84.000	1.073	
5	0.600	93.000	93.000	-1.341	
6	1.211	81.000	81.000	1.877	

Bonferroni T table value = 2.51   (1 Tailed Value, P=0.05, df=22,5)

d36 survival
File: 6114d36   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

| | | NUM OF | Minimum Sig Diff | % of | DIFFERENCE |
GROUP	IDENTIFICATION	REPS	(IN ORIG. UNITS)	CONTROL	FROM CONTROL
1	GRPS 1&2 POOLED	8			
2	0.075	4	9.357	10.6	-3.000
3	0.138	4	9.357	10.6	7.000
4	0.274	4	9.357	10.6	4.000
5	0.600	4	9.357	10.6	-5.000
6	1.211	4	9.357	10.6	7.000

----

d36 survival
File: 6114d36   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

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GROUP	ORIGINAL	TRANSFORMED	ISOTONIZED
IDENTIFICATION	N	MEAN	MEAN	MEAN

	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	8	88.000	88.000	89.000
2	0.075	4	91.000	91.000	89.000
3	0.138	4	81.000	81.000	86.000
4	0.274	4	84.000	84.000	86.000
5	0.600	4	93.000	93.000	86.000
6	1.211	4	81.000	81.000	81.000

d36 survival
File: 6114d36   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	89.000				
0.075	89.000	0.268		1.72	k= 1, v=22
0.138	86.000	0.536		1.80	k= 2, v=22
0.274	86.000	0.536		1.83	k= 3, v=22
0.600	86.000	0.536		1.84	k= 4, v=22
1.211	81.000	1.877	*	1.85	k= 5, v=22

s = 6.090
Note: df used for table values are approximate when v > 20.
d146 surv
File: 6114146   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	62.750	12.550	1.060
Within (Error)	8	94.750	11.844	
Total	13	157.500		

Critical F value = 3.69 (0.05,5,8)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

d146 surv
File: 6114146   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

                      TRANSFORMED   MEAN CALCULATED IN
GROUP   IDENTIFICATION   MEAN   ORIGINAL UNITS   T STAT SIG
----- ------------------- ----------- ------------------ ------ ---
  1   GRPS 1&2 POOLED   99.250   99.250
  2   0.075   93.500   93.500   1.929
  3   0.138   98.500   98.500   0.252
  4   0.274   95.000   95.000   1.426
  5   0.600   98.500   98.500   0.252
  6   1.211   98.500   98.500   0.252

Bonferroni T table value = 2.90   (1 Tailed Value, P=0.05, df=8,5)

d146 surv
File: 6114146   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

                      NUM OF   Minimum Sig Diff % of   DIFFERENCE
GROUP   IDENTIFICATION   REPS   (IN ORIG. UNITS) CONTROL FROM CONTROL
----- -------------------- ------- ---------------- ------- ------------
  1   GRPS 1&2 POOLED   4
  2   0.075   2   8.634   8.7   5.750
  3   0.138   2   8.634   8.7   0.750
  4   0.274   2   8.634   8.7   4.250
  5   0.600   2   8.634   8.7   0.750
  6   1.211   2   8.634   8.7   0.750
----

d146 surv
File: 6114146   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP   ORIGINAL   TRANSFORMED ISOTONIZED
        IDENTIFICATION   N   MEAN   MEAN   MEAN
------ -------------------- --- ----------- ----------- -----------
  1   GRPS 1&2 POOLED 4   99.250   99.250   99.250
  2   0.075 2   93.500   93.500   96.800
  3   0.138 2   98.500   98.500   96.800
  4   0.274 2   95.000   95.000   96.800
  5   0.600 2   98.500   98.500   96.800

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```
      6          1.211  2    98.500     98.500     96.800
```
----

d146 surv
File: 6114146   Transform: NO TRANSFORMATION

     WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	99.250				
0.075	96.800	0.822	1.86	k= 1, v= 8	
0.138	96.800	0.822	1.96	k= 2, v= 8	
0.274	96.800	0.822	2.00	k= 3, v= 8	
0.600	96.800	0.822	2.01	k= 4, v= 8	
1.211	96.800	0.822	2.02	k= 5, v= 8	

----
s =   3.441
Note: df used for table values are approximate when v > 20.

d237 % survival
File: 6114237   Transform: NO TRANSFORM

     KRUSKAL-WALLIS ANOVA BY RANKS - TABLE 1 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	RANK SUM
1	GRPS 1&2 POOLED	95.750	95.750	40.000
2	0.075	96.000	96.000	19.000
3	0.138	75.000	75.000	4.000
4	0.274	96.000	96.000	19.000
5	0.600	83.500	83.500	9.000
6	1.211	92.000	92.000	14.000

Calculated H Value = 7.591   Critical H Value Table = 11.070
Since Calc H < Crit H FAIL TO REJECT Ho:All groups are equal.

d237 % survival
File: 6114237   Transform: NO TRANSFORM

DUNNS MULTIPLE COMPARISON - KRUSKAL-WALLIS - TABLE 2 OF 2

                        GROUP

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```
                  TRANSFORMED  ORIGINAL  0 0 0 0 0 0
GROUP IDENTIFICATION   MEAN      MEAN    3 5 6 1 2 4
----- ---------------- --------  -------- - - - - - -
  3        0.138       75.000    75.000 \
  5        0.600       83.500    83.500 . \
  6        1.211       92.000    92.000 . . \
  1   GRPS 1&2 POOLED            95.750   95.750 . . . \
  2        0.075       96.000    96.000 . . . . \
  4        0.274       96.000    96.000 . . . . . \
----------------------------------------------------------------------
```

* = significant difference (p=0.05)   . = no significant difference
Table q value (0.05,6) = 2.936   Unequal reps - multiple SE values

d254 % survival
File: 6114254   Transform: NO TRANSFORMATION

                    ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	636.429	127.286	0.927
Within (Error)	8	1098.500	137.313	
Total	13	1734.929		

Critical F value = 3.69 (0.05,5,8)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

d254 % survival
File: 6114254   Transform: NO TRANSFORMATION

    BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

		TRANSFORMED	MEAN CALCULATED IN		
GROUP	IDENTIFICATION	MEAN	ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	85.500	85.500		
2	0.075	83.500	83.500	0.197	
3	0.138	71.000	71.000	1.429	
4	0.274	96.000	96.000	-1.035	
5	0.600	83.500	83.500	0.197	
6	1.211	83.500	83.500	0.197	

Bonferroni T table value = 2.90   (1 Tailed Value, P=0.05, df=8,5)

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d254 % survival
File: 6114254   Transform: NO TRANSFORMATION

       BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment
----
                  NUM OF   Minimum Sig Diff % of   DIFFERENCE
GROUP   IDENTIFICATION   REPS   (IN ORIG. UNITS) CONTROL FROM CONTROL
----- -------------------- ------- ---------------- ------- ------------
   1   GRPS 1&2 POOLED   4
   2   0.075   2   29.399   34.4   2.000
   3   0.138   2   29.399   34.4   14.500
   4   0.274   2   29.399   34.4   -10.500
   5   0.600   2   29.399   34.4   2.000
   6   1.211   2   29.399   34.4   2.000
----

d254 % survival
File: 6114254   Transform: NO TRANSFORMATION

       WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP   ORIGINAL   TRANSFORMED ISOTONIZED
        IDENTIFICATION   N   MEAN   MEAN   MEAN
------ -------------------- --- ----------- ----------- -----------
   1   GRPS 1&2 POOLED 4   85.500   85.500   85.500
   2   0.075   2   83.500   83.500   83.500
   3   0.138   2   71.000   71.000   83.500
   4   0.274   2   96.000   96.000   83.500
   5   0.600   2   83.500   83.500   83.500
   6   1.211   2   83.500   83.500   83.500
----

d254 % survival
File: 6114254   Transform: NO TRANSFORMATION

       WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

             ISOTONIZED CALC.   SIG   TABLE   DEGREES OF
   IDENTIFICATION   MEAN   WILLIAMS P=.05 WILLIAMS   FREEDOM
-------------------- ---------- ----------- ----- ----------- ------------
   GRPS 1&2 POOLED   85.500
       0.075   83.500   0.197   1.86   k= 1, v= 8
       0.138   83.500   0.197   1.96   k= 2, v= 8

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```
          0.274   83.500   0.197        2.00    k= 3, v= 8
          0.600   83.500   0.197        2.01    k= 4, v= 8
          1.211   83.500   0.197        2.02    k= 5, v= 8
```

s = 11.718
Note: df used for table values are approximate when v > 20.

length d34
File: 6114134   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	3.664	0.733	1.378
Within (Error)	18	9.582	0.532	
Total	23	13.246		

Critical F value = 2.77 (0.05,5,18)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

length d34
File: 6114134   Transform: NO TRANSFORMATION

DUNNETTS TEST - TABLE 1 OF 2   Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	solvent control	20.725	20.725		
2	0.075	19.750	19.750	1.890	
3	0.138	19.900	19.900	1.600	
4	0.274	19.950	19.950	1.503	
5	0.600	19.575	19.575	2.230	
6	1.211	19.575	19.575	2.230	

Dunnett table value = 2.41   (1 Tailed Value, P=0.05, df=18,5)

length d34
File: 6114134   Transform: NO TRANSFORMATION

DUNNETTS TEST - TABLE 2 OF 2   Ho:Control<Treatment

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GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	solvent control	4			
2	0.075	4	1.243	6.0	0.975
3	0.138	4	1.243	6.0	0.825
4	0.274	4	1.243	6.0	0.775
5	0.600	4	1.243	6.0	1.150
6	1.211	4	1.243	6.0	1.150

length d34
File: 6114l34   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	solvent control	4	20.725	20.725	20.725
2	0.075	4	19.750	19.750	19.867
3	0.138	4	19.900	19.900	19.867
4	0.274	4	19.950	19.950	19.867
5	0.600	4	19.575	19.575	19.575
6	1.211	4	19.575	19.575	19.575

length d34
File: 6114l34   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG	TABLE P=.05	DEGREES OF WILLIAMS	FREEDOM
solvent control	20.725					
0.075	19.867	1.664		1.73	k= 1, v=18	
0.138	19.867	1.664		1.82	k= 2, v=18	
0.274	19.867	1.664		1.85	k= 3, v=18	
0.600	19.575	2.229	*	1.86	k= 4, v=18	
1.211	19.575	2.229	*	1.87	k= 5, v=18	

s =   0.730
Note: df used for table values are approximate when v > 20.

33

34

length d63
File: 6114163   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	51.285	10.257	8.115
Within (Error)	8	10.112	1.264	
Total	13	61.397		

Critical F value = 3.69 (0.05,5,8)
Since F > Critical F REJECT Ho:All groups equal

length d63
File: 6114163   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	36.425	36.425		
2	0.075	37.200	37.200	-0.796	
3	0.138	38.200	38.200	-1.823	
4	0.274	35.800	35.800	0.642	
5	**0.600**	**35.700**	**35.700**	**0.745**	
6	1.211	31.650	31.650	4.904	*

Bonferroni T table value = 2.90   (1 Tailed Value, P=0.05, df=8,5)

length d63
File: 6114163   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.075	2	2.821	7.7	-0.775
3	0.138	2	2.821	7.7	-1.775

4	0.274	2	2.821	7.7	0.625
5	0.600	2	2.821	7.7	0.725
6	1.211	2	2.821	7.7	4.775

---

length d63
File: 6114163   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP IDENTIFICATION		N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	4	36.425	36.425	37.063
2	0.075	2	37.200	37.200	37.063
3	0.138	2	38.200	38.200	37.063
4	0.274	2	35.800	35.800	35.800
5	0.600	2	35.700	35.700	35.700
6	1.211	2	31.650	31.650	31.650

---

length d63
File: 6114163   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	37.063				
0.075	37.063	0.655		1.86	$k=1, v=8$
0.138	37.063	0.655		1.96	$k=2, v=8$
0.274	35.800	0.642		2.00	$k=3, v=8$
0.600	35.700	0.745		2.01	$k=4, v=8$
1.211	31.650	4.904	*	2.02	$k=5, v=8$

$s = 1.124$
Note: df used for table values are approximate when $v > 20$.

female length
File: 6114fl   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F

35

```
--------------------------------------------------------------------
Between        5        8.594        1.719        2.006

Within (Error)  8        6.860        0.857
--------------------------------------------------------------------
Total         13       15.454
--------------------------------------------------------------------
```

Critical F value = 3.69 (0.05,5,8)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

female length
File: 6114fl   Transform: NO TRANSFORMATION

    BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

		TRANSFORMED	MEAN CALCULATED IN		
GROUP	IDENTIFICATION	MEAN	ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	45.550	45.550		
2	0.075 46.300		46.300	-0.935	
3	0.138 47.800		47.800	-2.806	
4	0.274 45.950		45.950	-0.499	
5	0.600 45.900		45.900	-0.437	
6	1.211 45.250		45.250	0.374	

Bonferroni T table value = 2.90   (1 Tailed Value, P=0.05, df=8,5)

female length
File: 6114fl   Transform: NO TRANSFORMATION

    BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

		NUM OF	Minimum Sig Diff	% of	DIFFERENCE
GROUP	IDENTIFICATION	REPS	(IN ORIG. UNITS)	CONTROL	FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.075	2	2.323	5.1	-0.750
3	0.138	2	2.323	5.1	-2.250
4	0.274	2	2.323	5.1	-0.400
5	0.600	2	2.323	5.1	-0.350
6	1.211	2	2.323	5.1	0.300

female length
File: 6114fl   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP IDENTIFICATION		N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	4	45.550	45.550	46.300
2	0.075	2	46.300	46.300	46.300
3	0.138	2	47.800	47.800	46.300
4	0.274	2	45.950	45.950	45.950
5	0.600	2	45.900	45.900	45.900
6	1.211	2	45.250	45.250	45.250

female length
File: 6114fl   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	46.300				
0.075	46.300	0.935	1.86	k= 1, v= 8	
0.138	46.300	0.935	1.96	k= 2, v= 8	
0.274	45.950	0.499	2.00	k= 3, v= 8	
0.600	45.900	0.436	2.01	k= 4, v= 8	
1.211	45.250	0.374	2.02	k= 5, v= 8	

s = 0.926
Note: df used for table values are approximate when v > 20.

female weight
File: 6114fw   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.481	0.096	3.556
Within (Error)	8	0.214	0.027	
Total	13	0.694		

Critical F value = 3.69 (0.05,5,8)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

female weight
File: 6114fw   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

| | | TRANSFORMED | MEAN CALCULATED IN | | |
GROUP	IDENTIFICATION	MEAN	ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	2.303	2.303		
2	0.075	2.087	2.087	1.518	
3	0.138	2.354	2.354	-0.357	
4	0.274	2.025	2.025	1.951	
5	0.600	2.005	2.005	2.096	
6	1.211	1.811	1.811	3.461	*

Bonferroni T table value = 2.90   (1 Tailed Value, P=0.05, df=8,5)

female weight
File: 6114fw   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

| | | NUM OF | Minimum Sig Diff | % of | DIFFERENCE |
GROUP	IDENTIFICATION	REPS	(IN ORIG. UNITS)	CONTROL	FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.075	2	0.412	17.9	0.216
3	0.138	2	0.412	17.9	-0.051
4	0.274	2	0.412	17.9	0.278
5	0.600	2	0.412	17.9	0.298
6	1.211	2	0.412	17.9	0.493

----

female weight
File: 6114fw   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

| GROUP | | ORIGINAL | TRANSFORMED | ISOTONIZED |
IDENTIFICATION	N	MEAN	MEAN	MEAN

1	GRPS 1&2 POOLED	4	2.303	2.303	2.303
2	0.075	2	2.087	2.087	2.220
3	0.138	2	2.354	2.354	2.220
4	0.274	2	2.025	2.025	2.025
5	0.600	2	2.005	2.005	2.005
6	1.211	2	1.811	1.811	1.811

female weight
File: 6114fw   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG	TABLE P=.05 WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	2.303				
0.075	2.220	0.584		1.86	$k=1, v=8$
0.138	2.220	0.584		1.96	$k=2, v=8$
0.274	2.025	1.962		2.00	$k=3, v=8$
0.600	2.005	2.108	*	2.01	$k=4, v=8$
1.211	1.811	3.481	*	2.02	$k=5, v=8$

$s =$ 0.163
Note: df used for table values are approximate when $v > 20$.

male length
File: 6114ml   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	58.207	11.641	3.712
Within (Error)	8	25.085	3.136	
Total	13	83.292		

Critical F value = 3.69 (0.05,5,8)
Since F > Critical F REJECT Ho:All groups equal

male length

39

40

File: 6114ml   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

		TRANSFORMED MEAN	CALCULATED IN ORIGINAL UNITS	T STAT	SIG
GROUP	IDENTIFICATION	MEAN			
1	GRPS 1&2 POOLED	57.800	57.800		
2	0.075	58.550	58.550	-0.489	
3	0.138	57.150	57.150	0.424	
4	**0.274**	**57.050**	**57.050**	**0.489**	
5	0.600	52.650	52.650	3.358	*
6	1.211	54.050	54.050	2.445	

Bonferroni T table value = 2.90   (1 Tailed Value, P=0.05, df=8,5)

male length
File: 6114ml   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

		NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
GROUP	IDENTIFICATION				
1	GRPS 1&2 POOLED	4			
2	0.075	2	4.443	7.7	-0.750
3	0.138	2	4.443	7.7	0.650
4	0.274	2	4.443	7.7	0.750
5	0.600	2	4.443	7.7	5.150
6	1.211	2	4.443	7.7	3.750

---

male length
File: 6114ml   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP		N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
	IDENTIFICATION				
1	GRPS 1&2 POOLED	4	57.800	57.800	58.050
2	0.075	2	58.550	58.550	58.050
3	0.138	2	57.150	57.150	57.150
4	0.274	2	57.050	57.050	57.050
5	0.600	2	52.650	52.650	53.350
6	1.211	2	54.050	54.050	53.350

----

male length
File: 6114ml   Transform: NO TRANSFORMATION

        WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

                ISOTONIZED CALC.   SIG   TABLE   DEGREES OF
IDENTIFICATION   MEAN   WILLIAMS P=.05 WILLIAMS   FREEDOM

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG	TABLE P=.05 WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	58.050				
0.075	58.050	0.163		1.86	k= 1, v= 8
0.138	57.150	0.424		1.96	k= 2, v= 8
0.274	57.050	0.489		2.00	k= 3, v= 8
0.600	53.350	2.902	*	2.01	k= 4, v= 8
1.211	53.350	2.902	*	2.02	k= 5, v= 8

----
s =   1.771
Note: df used for table values are approximate when v > 20.

B
File: 6114mw   Transform: NO TRANSFORMATION

                    ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	6.502	1.300	6.701
Within (Error)	6	1.163	0.194	
Total	11	7.665		

  Critical F value = 4.39 (0.05,5,6)
  Since F > Critical F REJECT Ho:All groups equal

male weight
File: 6114mw   Transform: NO TRANSFORMATION

    BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

               TRANSFORMED   MEAN CALCULATED IN
GROUP   IDENTIFICATION   MEAN   ORIGINAL UNITS   T STAT SIG

41

```
1    solvent control   5.815        5.815
2         0.075        5.076        5.076    1.677
3         0.138        4.674        4.674    2.591
4         0.274        4.245        4.245    3.563  *
5         0.600        3.938        3.938    4.260  *
6         1.211        3.605        3.605    5.017  *
```

Bonferroni T table value = 3.14   (1 Tailed Value, P=0.05, df=6,5)

male weight
File: 6114mw   Transform: NO TRANSFORMATION

    BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

                     NUM OF Minimum Sig Diff % of   DIFFERENCE
GROUP   IDENTIFICATION   REPS   (IN ORIG. UNITS) CONTROL FROM CONTROL

```
1    solvent control   2
2         0.075        2     1.384    23.8    0.739
3         0.138        2     1.384    23.8    1.141
4         0.274        2     1.384    23.8    1.570
5         0.600        2     1.384    23.8    1.877
6         1.211        2     1.384    23.8    2.210
```

male weight
File: 6114mw   Transform: NO TRANSFORMATION

    WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP   ORIGINAL   TRANSFORMED ISOTONIZED
        IDENTIFICATION   N   MEAN   MEAN   MEAN

```
1    solvent control  2   5.815       5.815      5.815
2         0.075       2   5.076       5.076      5.076
3         0.138       2   4.674       4.674      4.674
4         0.274       2   4.245       4.245      4.245
5         0.600       2   3.938       3.938      3.938
6         1.211       2   3.605       3.605      3.605
```

male weight
File: 6114mw   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

```
                ISOTONIZED  CALC.   SIG    TABLE   DEGREES OF
IDENTIFICATION    MEAN    WILLIAMS  P=.05  WILLIAMS  FREEDOM
```

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
solvent control	5.815				
0.075	**5.076**	**1.678**		**1.94**	**k= 1, v= 6**
0.138	4.674	2.592	*	2.06	k= 2, v= 6
0.274	4.245	3.565	*	2.10	k= 3, v= 6
0.600	3.938	4.262	*	2.12	k= 4, v= 6
1.211	3.605	5.019	*	2.13	k= 5, v= 6

s =   0.440
Note: df used for table values are approximate when v > 20.
egg production d148-237
File: 6114p1   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	10045480.464	2009096.093	1.750
Within (Error)	8	9185334.750	1148166.844	
Total	13	19230815.214		

Critical F value = 3.69 (0.05,5,8)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

egg production d148-237
File: 6114p1   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	4405.250	4405.250		
2	0.075	4713.500	4713.500	-0.332	
3	0.138	4661.500	4661.500	-0.276	
4	0.274	4185.500	4185.500	0.237	
5	0.600	3773.000	3773.000	0.681	
6	1.211	2084.500	2084.500	2.501	

Bonferroni T table value = 2.90   (1 Tailed Value, P=0.05, df=8,5)

egg production d148-237
File: 6114p1   Transform: NO TRANSFORMATION

BONFERRONI T-TEST   -   TABLE 2 OF 2   Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.075	2	2688.324	61.0	-308.250
3	0.138	2	2688.324	61.0	-256.250
4	0.274	2	2688.324	61.0	219.750
5	0.600	2	2688.324	61.0	632.250
6	1.211	2	2688.324	61.0	2320.750

----

egg production d148-237
File: 6114p1   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	4	4405.250	4405.250	4546.375
2	0.075	2	4713.500	4713.500	4546.375
3	0.138	2	4661.500	4661.500	4546.375
4	0.274	2	4185.500	4185.500	4185.500
5	0.600	2	3773.000	3773.000	3773.000
6	1.211	2	2084.500	2084.500	2084.500

----

egg production d148-237
File: 6114p1   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	4546.375				
0.075	4546.375	0.152		1.86	k= 1, v= 8
0.138	4546.375	0.152		1.96	k= 2, v= 8

44

```
0.274   4185.500   0.237        2.00    k= 3, v= 8
0.600   3773.000   0.681        2.01    k= 4, v= 8
1.211   2084.500   2.501   *    2.02    k= 5, v= 8
```
----

s = 1071.525
Note: df used for table values are approximate when v > 20.

egg production 148-254
File: 6114p2   Transform: NO TRANSFORMATION

                    ANOVA TABLE

SOURCE   DF   SS   MS   F

Between   5   18075309.429   3615061.886   2.717

Within (Error) 8   10642539.500   1330317.438

Total   13   28717848.929

Critical F value = 3.69 (0.05,5,8)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

egg production 148-254
File: 6114p2   Transform: NO TRANSFORMATION

       BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment
----
              TRANSFORMED   MEAN CALCULATED IN
GROUP   IDENTIFICATION   MEAN   ORIGINAL UNITS   T STAT SIG
----- ---------------   -----------   ------------------ ------ ---
  1   GRPS 1&2 POOLED 5175.000   5175.000
  2   0.075 5861.500   5861.500   -0.687
  3   0.138 5812.000   5812.000   -0.638
  4   0.274 4926.000   4926.000   0.249
  5   0.600 4506.500   4506.500   0.669
  6   1.211 2249.500   2249.500   2.929 *

Bonferroni T table value = 2.90   (1 Tailed Value, P=0.05, df=8,5)

egg production 148-254
File: 6114p2   Transform: NO TRANSFORMATION

       BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

                          45

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.075	2	2893.722	55.9	-686.500
3	0.138	2	2893.722	55.9	-637.000
4	0.274	2	2893.722	55.9	249.000
5	0.600	2	2893.722	55.9	668.500
6	1.211	2	2893.722	55.9	2925.500

egg production 148-254
File: 6114p2   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	4	5175.000	5175.000	5505.875
2	0.075	2	5861.500	5861.500	5505.875
3	0.138	2	5812.000	5812.000	5505.875
4	0.274	2	4926.000	4926.000	4926.000
5	0.600	2	4506.500	4506.500	4506.500
6	1.211	2	2249.500	2249.500	2249.500

egg production 148-254
File: 6114p2   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG	TABLE P=.05 WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	5505.875				
0.075	5505.875	0.331		1.86	$k=1, v=8$
0.138	5505.875	0.331		1.96	$k=2, v=8$
0.274	4926.000	0.249		2.00	$k=3, v=8$
0.600	4506.500	0.669		2.01	$k=4, v=8$
1.211	2249.500	2.929	*	2.02	$k=5, v=8$

$s = 1153.394$
Note: df used for table values are approximate when $v > 20$.

percent viable eggs
File: 6114ve   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	222.212	44.442	2.993
Within (Error)	8	118.798	14.850	
Total	13	341.009		

Critical F value = 3.69 (0.05,5,8)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

percent viable eggs
File: 6114ve   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

| | | TRANSFORMED | MEAN CALCULATED IN | | |
GROUP	IDENTIFICATION	MEAN	ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	87.825	87.825		
2	0.075 87.850		87.850	-0.007	
3	0.138 87.800		87.800	0.007	
4	0.274 89.500		89.500	-0.502	
5	0.600 83.300		83.300	1.356	
6	1.211 77.150		77.150	3.199	*

Bonferroni T table value = 2.90   (1 Tailed Value, P=0.05, df=8,5)

percent viable eggs
File: 6114ve   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

| | | NUM OF | Minimum Sig Diff | % of | DIFFERENCE |
GROUP	IDENTIFICATION	REPS	(IN ORIG. UNITS)	CONTROL	FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.075	2	9.668	11.0	-0.025
3	0.138	2	9.668	11.0	0.025

47

48

4	0.274	2	9.668	11.0	-1.675
5	0.600	2	9.668	11.0	4.525
6	1.211	2	9.668	11.0	10.675

percent viable eggs
File: 6114ve   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	4	87.825	87.825	88.160
2	0.075	2	87.850	87.850	88.160
3	0.138	2	87.800	87.800	88.160
4	0.274	2	89.500	89.500	88.160
5	0.600	2	83.300	83.300	83.300
6	1.211	2	77.150	77.150	77.150

percent viable eggs
File: 6114ve   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	88.160				
0.075	88.160	0.100		1.86	k= 1, v= 8
0.138	88.160	0.100		1.96	k= 2, v= 8
0.274	88.160	0.100		2.00	k= 3, v= 8
0.600	83.300	1.356		2.01	k= 4, v= 8
1.211	77.150	3.199	*	2.02	k= 5, v= 8

s = 3.854
Note: df used for table values are approximate when v > 20.

eggs/female
File: 6114ef   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F

48

49

```
------------------------------------------------------------------------
Between         5      3124461.857    624892.371      2.277

Within (Error)  8      2195023.000    274377.875
------------------------------------------------------------------------
Total          13      5319484.857
------------------------------------------------------------------------
```

Critical F value = 3.69 (0.05,5,8)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

eggs/female
File: 6114ef   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

```
------------------------------------------------------------------------
                  TRANSFORMED   MEAN CALCULATED IN
GROUP  IDENTIFICATION      MEAN      ORIGINAL UNITS   T STAT SIG
-----  ------------------  ----------  ---------------  ------ ---
  1    GRPS 1&2 POOLED   2202.500        2202.500
  2           0.075   2455.000          2455.000      -0.557
  3           0.138   2671.000          2671.000      -1.033
  4           0.274   2134.000          2134.000       0.151
  5           0.600   2096.000          2096.000       0.235
  6           1.211   1056.000          1056.000       2.527
------------------------------------------------------------------------
```
Bonferroni T table value = 2.90   (1 Tailed Value, P=0.05, df=8,5)

eggs/female
File: 6114ef   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

```
------------------------------------------------------------------------
                  NUM OF  Minimum Sig Diff % of   DIFFERENCE
GROUP  IDENTIFICATION   REPS    (IN ORIG. UNITS) CONTROL  FROM CONTROL
-----  ------------------  -------  ---------------  -------  -----------
  1    GRPS 1&2 POOLED    4
  2           0.075   2      1314.176      59.7    -252.500
  3           0.138   2      1314.176      59.7    -468.500
  4           0.274   2      1314.176      59.7      68.500
  5           0.600   2      1314.176      59.7     106.500
  6           1.211   2      1314.176      59.7    1146.500
------------------------------------------------------------------------
```

eggs/female
File: 6114ef   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	4	2202.500	2202.500	2382.750
2	0.075	2	2455.000	2455.000	2382.750
3	0.138	2	2671.000	2671.000	2382.750
4	0.274	2	2134.000	2134.000	2134.000
5	0.600	2	2096.000	2096.000	2096.000
6	1.211	2	1056.000	1056.000	1056.000

eggs/female
File: 6114ef   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	2382.750				
0.075	2382.750	0.397		1.86	k= 1, v= 8
0.138	2382.750	0.397		1.96	k= 2, v= 8
0.274	2134.000	0.151		2.00	k= 3, v= 8
0.600	2096.000	0.235		2.01	k= 4, v= 8
1.211	1056.000	2.527	*	2.02	k= 5, v= 8

s = 523.811
Note: df used for table values are approximate when v > 20.

eggs/spawn
File: 6114es   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	7031.179	1406.236	1.592
Within (Error)	8	7067.750	883.469	
Total	13	14098.929		

Critical F value = 3.69 (0.05,5,8)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

eggs/spawn
File: 6114es   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

                  TRANSFORMED   MEAN CALCULATED IN
GROUP IDENTIFICATION   MEAN   ORIGINAL UNITS   T STAT SIG
----- -------------------   ----------   ------------------- ------ ---

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	154.250	154.250		
2	0.075	143.000	143.000	0.437	
3	0.138	180.500	180.500	-1.020	
4	0.274	170.500	170.500	-0.631	
5	0.600	132.000	132.000	0.864	
6	1.211	108.000	108.000	1.797	

Bonferroni T table value = 2.90   (1 Tailed Value, P=0.05, df=8,5)

eggs/spawn
File: 6114es   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

               NUM OF   Minimum Sig Diff % of   DIFFERENCE
GROUP IDENTIFICATION   REPS   (IN ORIG. UNITS) CONTROL FROM CONTROL
----- -------------------- ------- ---------------- ------- -----------

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.075	2	74.572	48.3	11.250
3	0.138	2	74.572	48.3	-26.250
4	0.274	2	74.572	48.3	-16.250
5	0.600	2	74.572	48.3	22.250
6	1.211	2	74.572	48.3	46.250

----

eggs/spawn
File: 6114es   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP   ORIGINAL   TRANSFORMED   ISOTONIZED
      IDENTIFICATION   N   MEAN   MEAN   MEAN

```
------  --------------------- --- ----------  -----------  -----------
  1     GRPS 1&2 POOLED  4   154.250      154.250      160.500
  2         0.075  2   143.000      143.000      160.500
  3         0.138  2   180.500      180.500      160.500
  4         0.274  2   170.500      170.500      160.500
  5         0.600  2   132.000      132.000      132.000
  6         1.211  2   108.000      108.000      108.000
------------------------------------------------------------------
```

eggs/spawn
File: 6114es   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	160.500				
0.075	160.500	0.243	1.86	$k= 1, v= 8$	
0.138	160.500	0.243	1.96	$k= 2, v= 8$	
0.274	160.500	0.243	2.00	$k= 3, v= 8$	
0.600	132.000	0.864	2.01	$k= 4, v= 8$	
1.211	108.000	1.797	2.02	$k= 5, v= 8$	

$s = 29.723$
Note: df used for table values are approximate when $v > 20$.

spawns/female
File: 6114sf   Transform: NO TRANSFORMATION

KRUSKAL-WALLIS ANOVA BY RANKS - TABLE 1 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	RANK SUM
1	GRPS 1&2 POOLED	14.250	14.250	35.000
2	0.075	17.000	17.000	25.000
3	0.138	12.500	12.500	12.000
4	0.274	11.500	11.500	8.500
5	0.600	14.000	14.000	16.500
6	1.211	8.000	8.000	8.000

Calculated H Value = 6.281   Critical H Value Table = 11.070
Since Calc H < Crit H FAIL TO REJECT Ho:All groups are equal.

spawns/female
File: 6114sf   Transform: NO TRANSFORMATION

DUNNS MULTIPLE COMPARISON - KRUSKAL-WALLIS - TABLE 2 OF 2

```
----------------------------------------------------------------------------
                              GROUP
              TRANSFORMED  ORIGINAL  0 0 0 0 0 0
GROUP  IDENTIFICATION    MEAN      MEAN    6 4 3 5 1 2
----- ---------------- ----------- --------- - - - - - -
  6         1.211      8.000     8.000 \
  4         0.274     11.500    11.500 . \
  3         0.138     12.500    12.500 .. \
  5         0.600     14.000    14.000 ... \
  1    GRPS 1&2 POOLED    14.250    14.250 .... \
  2         0.075     17.000    17.000 ..... \
----------------------------------------------------------------------------
```

* = significant difference (p=0.05)   . = no significant difference
Table q value (0.05,6) = 2.936   Unequal reps - multiple SE values

F1 % hatch
File: 6114h2   Transform: NO TRANSFORMATION

KRUSKAL-WALLIS ANOVA BY RANKS - TABLE 1 OF 2

```
----------------------------------------------------------------------------
               TRANSFORMED  MEAN CALCULATED IN     RANK
GROUP  IDENTIFICATION     MEAN      ORIGINAL UNITS     SUM
----- -------------------- ----------- ----------------- -----------
  1    GRPS 1&2 POOLED   96.120       96.120    1319.000
  2         0.075    90.929       90.929     613.000
  3         0.138    93.083       93.083     490.000
  4         0.274    97.462       97.462     676.500
  5         0.600    91.333       91.333     400.500
  6         1.211    77.400       77.400     242.000
----------------------------------------------------------------------------
```

Calculated H Value = 4.845   Critical H Value Table = 11.070
Since Calc H < Crit H FAIL TO REJECT Ho:All groups are equal.

F1 % hatch
File: 6114h2   Transform: NO TRANSFORMATION

DUNNS MULTIPLE COMPARISON - KRUSKAL-WALLIS - TABLE 2 OF 2

```
----------------------------------------------------------------------------
                              GROUP
              TRANSFORMED  ORIGINAL  0 0 0 0 0 0
GROUP  IDENTIFICATION    MEAN      MEAN    6 2 5 3 1 4
----- ---------------- ----------- --------- - - - - - -
```

```
6        1.211    77.400    77.400 \
2        0.075    90.929    90.929 .\
5        0.600    91.333    91.333 ..\
3        0.138    93.083    93.083 ...\
1    GRPS 1&2 POOLED    96.120    96.120 *...\
4        0.274    97.462    97.462 .....\
```
----

* = significant difference (p=0.05)   . = no significant difference
Table q value (0.05,6) = 2.936   Unequal reps - multiple SE values

F1 egg and alevin survival
File: 6114s2   Transform: NO TRANSFORMATION

KRUSKAL-WALLIS ANOVA BY RANKS - TABLE 1 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	RANK SUM
1	GRPS 1&2 POOLED	92.240	92.240	1207.500
2	0.075	86.000	86.000	544.500
3	0.138	91.667	91.667	540.000
4	0.274	94.846	94.846	702.000
5	0.600	89.500	89.500	469.500
6	1.211	75.000	75.000	277.500

Calculated H Value = 5.834   Critical H Value Table = 11.070
Since Calc H < Crit H FAIL TO REJECT Ho:All groups are equal.

F1 egg and alevin survival
File: 6114s2   Transform: NO TRANSFORMATION

DUNNS MULTIPLE COMPARISON - KRUSKAL-WALLIS - TABLE 2 OF 2

```
                       GROUP
        TRANSFORMED  ORIGINAL  0 0 0 0 0 0
GROUP  IDENTIFICATION  MEAN     MEAN   6 2 5 3 1 4
```
----- --------------- ----------- --------- - - - - - -
```
6        1.211    75.000    75.000 \
2        0.075    86.000    86.000 .\
5        0.600    89.500    89.500 ..\
3        0.138    91.667    91.667 ...\
1    GRPS 1&2 POOLED    92.240    92.240 ....\
4        0.274    94.846    94.846 .....\
```
----

* = significant difference (p=0.05)   . = no significant difference

Table q value (0.05,6) = 2.936   Unequal reps - multiple SE values

b
File: 611435s   Transform: NO TRANSFORMATION

                        ANOVA TABLE

SOURCE   DF   SS   MS   F

Between   5   300.724   60.145   0.488

Within (Error) 39   4806.476   123.243

Total   44   5107.200

 Critical F value = 2.53 (0.05,5,30)
 Since F < Critical F FAIL TO REJECT Ho:All groups equal

F1 incubation d35 survivorship
File: 611435s   Transform: NO TRANSFORMATION

    BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

                TRANSFORMED   MEAN CALCULATED IN
GROUP IDENTIFICATION   MEAN   ORIGINAL UNITS   T STAT SIG
----
  1   GRPS 1&2 POOLED   87.143   87.143
  2   0.075 90.286   90.286   -0.612
  3   0.138 93.333   93.333   -1.143
  4   0.274 93.333   93.333   -1.143
  5   0.600 91.333   91.333   -0.774
  6   1.211 87.333   87.333   -0.035

Bonferroni T table value = 2.43   (1 Tailed Value, P=0.05, df=39,5)

F1 incubation d35 survivorship
File: 611435s   Transform: NO TRANSFORMATION

    BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

                NUM OF   Minimum Sig Diff % of   DIFFERENCE
GROUP   IDENTIFICATION   REPS   (IN ORIG. UNITS) CONTROL FROM CONTROL
----
  1   GRPS 1&2 POOLED   14

2	0.075	7	12.467	14.3	-3.143
3	0.138	6	13.142	15.1	-6.190
4	0.274	6	13.142	15.1	-6.190
5	0.600	6	13.142	15.1	-4.190
6	1.211	6	13.142	15.1	-0.190

F1 incubation d35 survivorship
File: 611435s   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP IDENTIFICATION	ORIGINAL N	MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1 GRPS 1&2 POOLED	14	87.143	87.143	87.143
2 0.075	7	90.286	90.286	90.286
3 0.138	6	93.333	93.333	91.333
4 0.274	6	93.333	93.333	91.333
5 0.600	6	91.333	91.333	91.333
6 1.211	6	87.333	87.333	91.333

F1 incubation d35 survivorship
File: 611435s   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	87.143				
0.075	90.286	0.612	1.69		k= 1, v=39
0.138	91.333	0.774	1.77		k= 2, v=39
0.274	91.333	0.774	1.79		k= 3, v=39
0.600	91.333	0.774	1.80		k= 4, v=39
1.211	91.333	0.774	1.81		k= 5, v=39

s = 11.101
Note: df used for table values are approximate when v > 20.

F1 length
File: 61141   Transform: NO TRANSFORMATION

ANOVA TABLE

56

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SOURCE	DF	SS	MS	F
Between	5	23.777	4.755	3.712
Within (Error)	39	49.971	1.281	
Total	44	73.748		

Critical F value = 2.53 (0.05,5,30)
Since F > Critical F REJECT Ho:All groups equal

F1 length
File: 61141   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	22.479	22.479		
2	0.075	22.257	22.257	0.423	
3	0.138	22.733	22.733	-0.461	
4	0.274	22.050	22.050	0.776	
5	0.600	21.933	21.933	0.987	
6	1.211	20.317	20.317	3.915	*

Bonferroni T table value = 2.43   (1 Tailed Value, P=0.05, df=39,5)

F1 length
File: 61141   Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	14			
2	0.075	7	1.271	5.7	0.221
3	0.138	6	1.340	6.0	-0.255
4	0.274	6	1.340	6.0	0.429
5	0.600	6	1.340	6.0	0.545
6	1.211	6	1.340	6.0	2.162

F1 length
File: 61141   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	14	22.479	22.479	22.479
2	0.075	7	22.257	22.257	22.477
3	0.138	6	22.733	22.733	22.477
4	0.274	6	22.050	22.050	22.050
5	0.600	6	21.933	21.933	21.933
6	1.211	6	20.317	20.317	20.317

F1 length
File: 61141   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	22.479				
0.075	22.477	0.003		1.69	k= 1, v=39
0.138	22.477	0.003		1.77	k= 2, v=39
0.274	22.050	0.776		1.79	k= 3, v=39
0.600	**21.933**	**0.987**		**1.80**	**k= 4, v=39**
1.211	20.317	3.914	*	1.81	k= 5, v=39

s =   1.132
Note: df used for table values are approximate when v > 20.

F1 weight
File: 6114w   Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	16305.778	3261.156	4.215
Within (Error)	39	30176.191	773.748	

Total   44   46481.970

Critical F value =   2.53 (0.05,5,30)
Since F > Critical F REJECT Ho:All groups equal

F1 weight
File: 6114w   Transform: NO TRANSFORMATION

     BONFERRONI T-TEST - TABLE 1 OF 2   Ho:Control<Treatment

                    TRANSFORMED   MEAN CALCULATED IN
GROUP   IDENTIFICATION   MEAN   ORIGINAL UNITS   T STAT SIG
----- -------------------- ----------- ----------------- ------ ---

1	GRPS 1&2 POOLED	217.836	217.836		
2	0.075	215.329	215.329	0.195	
3	0.138	214.200	214.200	0.268	
4	0.274	201.983	201.983	1.168	
5	0.600	202.417	202.417	1.136	
6	1.211	159.083	159.083	4.329	*

Bonferroni T table value = 2.43   (1 Tailed Value, P=0.05, df=39,5)

F1 weight
File: 6114w   Transform: NO TRANSFORMATION

     BONFERRONI T-TEST - TABLE 2 OF 2   Ho:Control<Treatment

                   NUM OF Minimum Sig Diff % of   DIFFERENCE
GROUP   IDENTIFICATION   REPS   (IN ORIG. UNITS) CONTROL FROM CONTROL
----- -------------------- ------- ---------------- ------- -----------

1	GRPS 1&2 POOLED	14			
2	0.075	7	31.238	14.3	2.507
3	0.138	6	32.928	15.1	3.636
4	0.274	6	32.928	15.1	15.852
5	0.600	6	32.928	15.1	15.419
6	1.211	6	32.928	15.1	58.752

F1 weight
File: 6114w   Transform: NO TRANSFORMATION

     WILLIAMS TEST (Isotonic regression model)   TABLE 1 OF 2

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GROUP			ORIGINAL	TRANSFORMED	ISOTONIZED
	IDENTIFICATION	N	MEAN	MEAN	MEAN
1	GRPS 1&2 POOLED	14	217.836	217.836	217.836
2	0.075	7	215.329	215.329	215.329
3	0.138	6	214.200	214.200	214.200
4	0.274	6	201.983	201.983	202.200
5	0.600	6	202.417	202.417	202.200
6	1.211	6	159.083	159.083	159.083

F1 weight
File: 6114w   Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model)   TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG	TABLE WILLIAMS P=.05	DEGREES OF FREEDOM
GRPS 1&2 POOLED	217.836				
0.075	215.329	0.195		1.69	k= 1, v=39
0.138	214.200	0.268		1.77	k= 2, v=39
0.274	202.200	1.152		1.79	k= 3, v=39
0.600	**202.200**	**1.152**		**1.80**	**k= 4, v=39**
1.211	159.083	4.329	*	1.81	k= 5, v=39

$s = 27.816$
Note: df used for table values are approximate when $v > 20$.